REMARKS

Claims 25-36 are all the claims pending in the application.

Claims 25-36 have been amended to remove specific reference to the apparatus claimed therein as either a "measuring" apparatus as in claim 25, or as a "quantitative" apparatus as in claims 26-35. Claim 25 specifically states that the apparatus is "for measuring a value." Thus, the inclusion of the term "measuring" or quantitative" in the preamble is only redundant.

Claim 25 has also been amended to more clearly refer to the measuring being "corrected for" rather than "based on" a value detected from the first label. Support for this change can be found in the last line of the claim where it states that the "quantity of said organism-originated substance bound to said specific binding substance, based on the detected level of said second labeling signal, corrected for the detected level of said first labeling signal."

Claim 26, 27 and 28 have been amended to correct obvious grammatical errors.

Claims 27 and 28 have also been amended to more clearly state that the characteristic values are "corrected values" as first recited in independent claim 25. Support for this amendment may be found at page 13, line 19 – page 15, line 3.

Thus, no new matter has been added and entry of the Amendment is earnestly solicited.

I. Objection of claims under 37 C.F.R. §1.75(c)

At page 2 of the Office Action, paragraph 2, claims 28, 31, 32, 35 and 36 are objected to as improper dependent claims under 37 C.F.R. §1.75(c).

The Examiner alleges that the cited claims fail to further limit the subject matter of the claims from which they depend. Specifically, the Examiner contends that claim 28 does not

further limit claim 26, claims 31 and 35 do not further limit claim 27, and claims 32 and 36 do not further limit claim 28.

A. Claim 28

In response, Applicant asserts that claim 28 further limits the inventive apparatus recited in claim 26 by defining the means of performing the quantitative analysis. Therefore, in contrast to the Examiner's rejection, claim 28 does in fact set forth a further limitation to claim 26.

However, in order to further the prosecution of the application, Applicant has amended claim 28 to more clearly recite the additional limitation recited in claim 28. Applicant has amended the claim to describe the correction value as a value "calculated from information about 1) the length of the cDNA polynucleotide and 2) the relative frequency of the first labeling substance within each cDNA polynucleotide used in the method." Support for this amendment may be found at page 13, line 19 – page 15, line 3.

Accordingly, Applicant respectfully requests reconsideration and withdrawal of this portion of the objection.

B. Claims 31, 32, 35 and 36

Contrary to the Examiner's position, Applicant asserts that each of claims 31 and 35 clearly further limit claim 27. Claim 27 depends from claim 25, and claim 25 merely recites a "first labeling substance." Claim 31 further defines the first labeling substance as a fluorescent dye and claim 35 further defines it as a radioactive isotope.

Similarly, Applicant asserts that each of claims 32 and 36 clearly further limit claim 28. Claim 28 depends from claim 26, which in turn depends from claim 25, and claim 25 merely

recites a "first labeling substance." Claim 32 further defines the first labeling substance as a fluorescent dye and claim 36 further defines it as a radioactive isotope.

Thus, in contrast to the Examiner's position, claims 31 and 35 further limit claim 27, and claims 32 and 36 further limit claim 28. Accordingly, Applicant respectfully requests reconsideration and withdrawal of this portion of the objection.

II. Rejection of claims under 35 U.S.C. §112

A. Claim 25

At page 3 of the Office Action, paragraph 4, claim 25-36 are rejected as being indefinite.

The Examiner is unclear as to meaning in claim 25 of the phrase "for measuring a value detected from a second label based on a value detected from a first label."

In response, Applicant has amended claim 25 to more clearly recite the invention. Amended claim 25 states that the value detected from the second label is "corrected for" based on the value detected from the first label. Thus, it is now clear that the value detected from a first label serves to take into account and correct for, or control for, variations in the quantity of specific binding substances (detected by the first label) disposed on the carrier.

Therefore, Applicant respectfully requests reconsideration and withdrawal of this portion of the rejection.

B. Claim 27

At page 3 of the Office Action, paragraph 5, claim 27 is rejected as being indefinite.

The Examiner claims that there is insufficient antecedent basis for "cDNA polynucleotides" in the claim.

In response, Applicant has amended claim 27 to recite "specific binding substance" in place of "cDNA polynucleotides."

Therefore, Applicant respectfully requests reconsideration and withdrawal of this portion of the rejection.

III. Rejection of claims under 35 U.S.C. § 102

At page 3 of the Office Action, paragraph 7, claims 25 and 29 are rejected under 35 U.S.C. §102(b) as being anticipated by Larin et al. (*Nuc. Acid Res.*, 1994).

The Examiner asserts that Larin et al. teaches a method of analyzing multiple probes by FISH to metaphase chromosomes using two different labeled signals and an imager system as an analyzing means. The Examiner contends that the propidium iodide (PI) counterstain qualifies as the first labeled signal, the fluorescent biotin-avidin complex as the second labeled signal, and the BioRAD optical system as the analyzing means. Thus, the Examiner asserts that Larin et al. teaches all of the limitations of claims 25 and 29.

In response, Applicant asserts that Larin et al. does not teach each of the limitations of claims 25 and 29. Specifically, Larin et al. does not provide a "a first detection means for detecting a level of a first labeling signal." The Examiner considers the PI counterstain a first labeled signal. Accepting that contention, Applicant notes that PI is an intercalating agent, inserting between the bases of DNA, and fluorescing under ultraviolet light. In FISH, the PI is needed to observe and identify the specific chromosomes under analysis.

However, even if the PI is considered a first labeled signal, the system described in Larin et al. does not provides a means for detecting a level of PI. There is no evidence in Larin et al.

that the BioRAD optical system measures any quantitative difference in the level of fluorescence emitted by the PI. The general staining function of PI is simply to assist in the karyotype determination of the metaphase chromosome spread.

In addition, Applicant asserts that Larin et al. does not teach the structural feature of an analyzing means for measuring the quantity of a substance based on the level of one signal corrected for by the value of a different signal.

Instead, Larin et al. teaches a method of FISH analysis which consists entirely of optically observing and manually counting the signals generated by the hybridization of centromere and YAC probes to metaphase chromosomes. The PI counterstain is used in Larin et al. to produce a contrasting red color and functions to discriminate particular metaphase chromosome spreads which the observer then determines are optimal for analysis. Larin et al. does not disclose a means for measuring the quantity of a substance based on the level of one signal corrected for by the value of a different signal. Larin et al. does not determine a quantitative measurement of the PI stain. The chromosomes are simply stained with PI in order to coordinate the identification of appropriate metaphase spreads and to verify the identity of particular chromosomes in which the specific probes are bound.

Thus again, Applicant asserts that Larin et al. does not teach each of the limitations of claims 25 and 29, and respectfully requests reconsideration and withdrawal of this rejection.

IV. Rejection of claims under 35 U.S.C. § 103

A. Claims 25 and 33

At page 4 of the Office Action, paragraph 9, claims 25 and 33 are rejected under 35 U.S.C. §103(a) as obvious over Arnold et al. (Mol. Endocrinol., 1995) in view of Brown et al. (U.S. Pat. No. 5,849,920) ("Brown '920").

The Examiner states that Arnold et al. teaches protein analysis by radioactive labeling, chemiluminescence, and x-ray film detection, and that Brown '920 reports the use of a densitometer for scanning x-ray films. The Examiner contends that it would have been *prima facie* obvious to one having ordinary skill in the art to modify and combine the teachings of the two methods and construct Applicant's claimed apparatus.

The Examiner contends that the ^{32}P-labeled hER qualifies as the "first labeling signal with known different specific binding substances," while the ECL visualization of hER qualifies as the "second labeled signal." The Examiner acknowledges that the Arnold et al. reference does not disclose an apparatus to scan an x-ray film.

In response, Applicant asserts that claims 25 and 33 are not obvious over Arnold et al., in view of Brown '920, for the following reasons.

First, Applicant states that, as admitted by the Examiner, neither of the two disclosures alone teaches an apparatus comprising a first detection means, a second detection means and an analyzing means for measuring the quantity of a substance based on the level of one signal corrected for by the value of a different signal. Instead, the Examiner contends that it would

have been obvious to combine the apparatus of Arnold et al. with that of Brown '920 to arrive at the present invention.

Applicant asserts, however, that the combination of these two disclosures does not result in the apparatus of the present invention.

First, there are not two different detection means in the disclosures of Arnold et al. and Brown '920, as there are in the apparatus of the present invention. Instead, both would rely on the same apparatus, i.e., one that scans an x-ray film.

Second, the x-ray film is produced by exposing the film to a light source (either produced chemically through ECL or radioactively through ^{32}P). In both cases, the x-ray film detects the production of light by forming a "spot" on the surface of the developed film. Thus, the detection means of the cited art measures the same signal (presence and size of spot). In contrast, the apparatus of the present invention utilizes detection means that measure two different signals ("a first labeling signal" and a "second labeling signal").

Third, while the Examiner states that the correction for the background could be considered to be "neglect", Applicant notes that if the initial ^{32}P-generated light source is significantly strong, it can interfere with the second ECL generated light source. Arnold et al. does not provide a means to discriminate between these two light sources. Both ECL-generated light source and the ^{32}P-generated light source are detected by a single means, that is x-ray film.

Finally, Applicant contends that the method of Arnold et al. describes procedures that are heavily dependant on manual intervention. Thus, the combination of the two references as the Examiner suggests does not lend itself to the type of automatic operation disclosed by the

Applicant's invention. Arnold et al. describes SDS-gel electrophoresis, western blot, autoradiographic detection of a signal generated from a ^{32}P-labeled phosphoprotein, followed by detection of signal generated using antibodies and ECL, all involving significant manual operation. Arnold et al. discloses further manual procedure by stripping the first probe from the western blot membrane and reprobing with a second probe.

Thus again, Applicant asserts that the teachings of Arnold et al., combined with the teachings of Brown '920, would not make obvious, or even suggest, the present invention. Accordingly, Applicant respectfully requests reconsideration and withdrawal of this rejection.

B. Claims 25-32.

At page 6 of the Office Action, paragraph 10, claims 25-32 are rejected under 35 U.S.C. §103(a) as being unpatentable over Brown et al., (U.S. Pat. No. 5,807,522; "Brown '522") in view of Cardullo et al. (PNAS, 1988).

The Examiner asserts that Brown '522 teaches cDNA microarrays hybridized with dual-labeled nucleic acid probes and that Cardullo et al. teaches a method and apparatus for the detection of nucleic acid hybridization. The Examiner contends it would have been obvious to combine the teachings of the two in order to produce the present claimed invention.

The Examiner acknowledges that neither Brown '522 nor Cardullo et al. teaches fluorescence labeled cDNA immobilized on a solid support (corresponding to Applicant's claim limitation of a first labeled binding substance disposed on a carrier). However, the Examiner alleges that it would have been obvious to construct a system whereby fluorescence-labeled cDNA is substituted for unlabeled cDNA in the Brown '522 microarray, and then combined with

the FRET technique of Cardullo et al. to detect and measure a hybridization signal (from a second labeled substance).

In response, Applicant asserts that the subject matter recited in claims 25-32 would not have been obvious over the teachings of Brown '522 in view of Cardullo et al. for the following reasons.

Applicant first asserts that it is not obvious to replace the detection method of Brown '522 with the method of Cardullo et al. Indeed, neither of the two references provide any suggestion to use FRET with nucleic acid fixed to a solid support. The FRET technique disclosed in Cardullo et al. is performed entirely in solution hybridization studies. At page 8790, Cardullo et al. state: "We report here that FRET provides a useful means for detection of nucleic acid hybridization in solution." In FRET, the hybridization between complementary strands of DNA takes place in solution in a cuvette. Hybridization is measured with a spectrophotometer by following changes in fluorescence resulting from proximity-induced dipole interactions between two fluorophores. There is no suggestion in Cardullo et al. that FRET could be adapted to measure hybridization of nucleic acid on a solid support.

Second, Cardullo et al. teaches away from the application of FRET to measure nucleic acid hybridization as disclosed by the present invention. The signals generated during hybridization by the dual fluorescence labels are highly temperature sensitive. At page 8792 and in Fig. 4 of Cardullo et al., the authors disclose the effects of temperature on fluorescence intensity. The data indicated that the measured values for rhodamine enhancement and fluorescein quenching were highly variable over a relatively narrow temperature range. One

skilled in the art would recognize the difficulty in adapting FRET to the analysis of nucleic acid hybridization while still obtaining the type of accurate and quantitative measurement that the present invention demands.

Furthermore, at page 8792, Cardullo et al. states: "On the basis of these studies and our experience so far, it is not apparent that FRET offers increased sensitivity over existing methods for detecting nucleic acid hybridization." The authors then go on to suggest that FRET could be useful for detecting qualitative hybridization *in vivo*, such as the observation of viral infection and replication in living cells. One skilled in the art would therefore find no motivation to adapt FRET to the quantitative analysis of hybridization as claimed by the present invention.

Thus, Applicant asserts that there is no motivation to substitute fluorescence-labeled cDNA for unlabeled cDNA in Brown '522 microarray. One skilled in the art would not expect that fluorescence-labeled cDNA fixed to a carrier would perform as the Examiner contends in a FRET type of analysis.

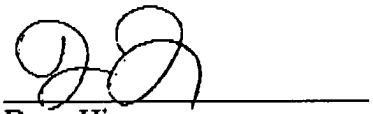
Therefore again, Applicant asserts that the subject matter recited in claims 25-32 would not have been obvious over the teachings or suggestions of Brown '522 in view of Cardullo et al. Accordingly, Applicant respectfully requests reconsideration and withdrawal of this rejection.

V. Conclusion

In view of the above, reconsideration and allowance of this application are now believed to be in order, and such actions are hereby solicited. If any points remain in issue which the Examiner feels may be best resolved through a personal or telephone interview, the Examiner is kindly requested to contact the undersigned at the telephone number listed below.

Applicant hereby petitions for any extension of time which may be required to maintain the pendency of this case, and any required fee, except for the Issue Fee, for such extension is to be charged to Deposit Account No. 19-4880.

Respectfully submitted,



Drew Hissong
Registration No. 44,765

SUGHRUE MION, PLLC
2100 Pennsylvania Avenue, N.W.
Washington, D.C. 20037-3213
Telephone: (202) 293-7060
Facsimile: (202) 293-7860

Date: April 4, 2002

APPENDIX

VERSION WITH MARKINGS TO SHOW CHANGES MADE

IN THE CLAIMS:

The claims are amended as follows:

25. (Twice amended) An ~~A measuring~~ apparatus for measuring a value detected from a second label corrected for ~~based on~~ a value detected from a first label comprising:

a first detection means for detecting a level of a first labeling signal emitted by a first labeling substance, which labels a plurality of known different specific binding substances respectively disposed at a plurality of predetermined positions on a carrier of a test piece, for each of said plurality of predetermined positions;

a second detection means for detecting a level of a second labeling signal emitted by a second labeling substance, which differs from said first labeling substance and labels an organism-originated substance bound to said specific binding substance, for each of said plurality of predetermined positions; and

an analyzing means for measuring a quantity of said organism-originated substance bound to said specific binding substance, based on the detected level of said second labeling signal, corrected for the detected level of said first labeling signal.

26. (Twice amended) The ~~quantitative~~ apparatus as set forth in claim 25, wherein said specific binding substance is a cDNA polynucleotide~~substances are cDNA polynucleotides~~.

27. (Twice amended) The ~~quantitative~~ apparatus as set forth in claim 25, wherein said analyzing means performing ~~further performs~~ said measurement, utilizes a correction value

calculated for each specific binding substance used in the method~~based on a characteristic value related to cDNA polynucleotides~~.

28. (Twice amended) The ~~quantitative~~ apparatus as set forth in claim 26, wherein said analyzing means performing ~~further performs~~ said measurement~~,~~ utilizes a correction value calculated from information about 1) the length of the cDNA polynucleotide and 2) the relative frequency of the first labeling substance within each cDNA polynucleotide used in the method~~based on a characteristic value related to cDNA polynucleotides~~.

29. (amended) The ~~quantitative~~ apparatus as set forth in claim 25, wherein said first labeling substance for said specific binding substances is a fluorescent dye.

30. (amended) The ~~quantitative~~ apparatus as set forth in claim 26, wherein said first labeling substance for said specific binding substances is a fluorescent dye.

31. (amended) The ~~quantitative~~ apparatus as set forth in claim 27, wherein said first labeling substance for said specific binding substances is a fluorescent dye.

32. (amended) The ~~quantitative~~ apparatus as set forth in claim 28, wherein said first labeling substance for said specific binding substances is a fluorescent dye.

33. (amended) The ~~quantitative~~ apparatus as set forth in claim 25, wherein said first labeling substance for said specific binding substances is a radioactive isotope.

34. (amended) The ~~quantitative~~ apparatus as set forth in claim 26, wherein said first labeling substance for said specific binding substances is a radioactive isotope.

35. (amended) The ~~quantitative~~ apparatus as set forth in claim 27, wherein said first labeling substance for said specific binding substances is a radioactive isotope.

36. (amended) The ~~quantitative~~ apparatus as set forth in claim 28, wherein said first labeling substance for said specific binding substances is a radioactive isotope.